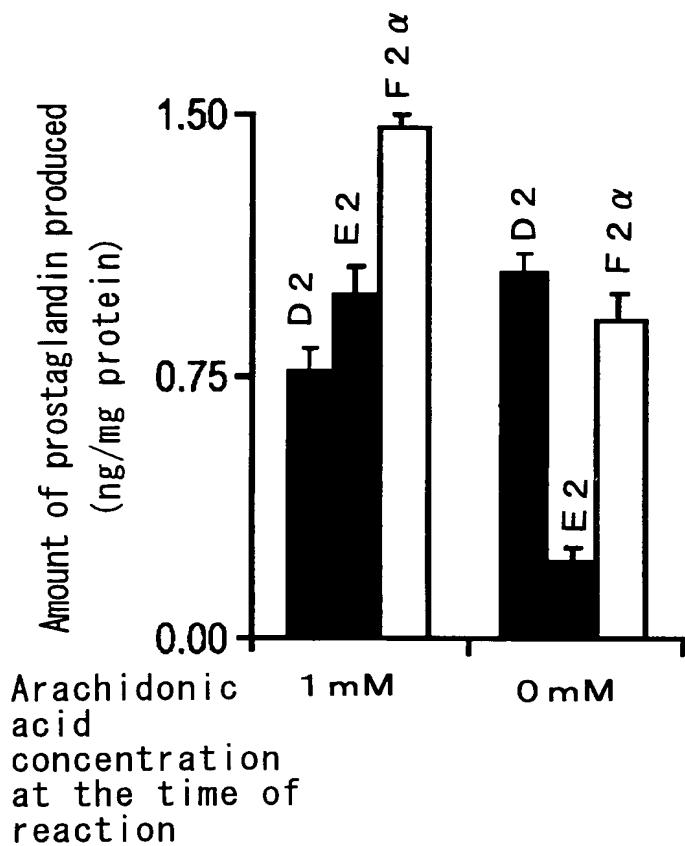
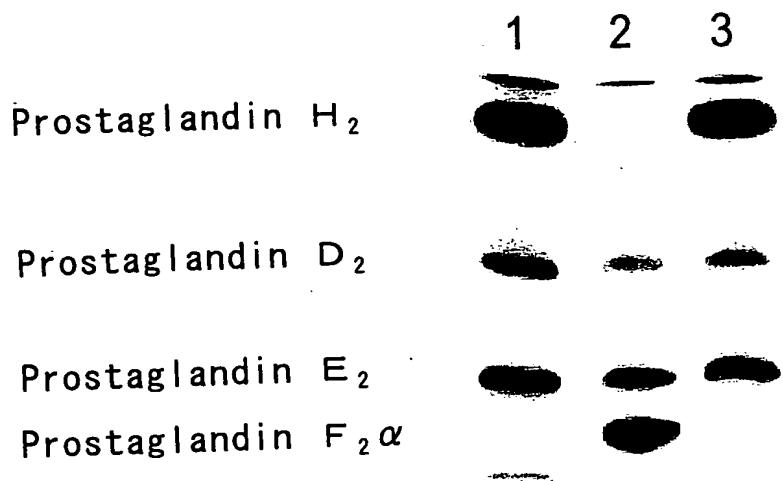


Fig. 1



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Fig. 2

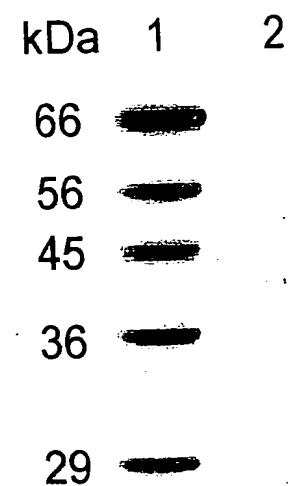


1. No enzyme
2. *Trypanosoma cruzi* extract
3. *Trypanosoma cruzi* extract after
heat treatment

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Fig. 3



1. Molecular-weight marker protein
2. Purified enzyme

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Fig. 4

Purification step	Total protein (mg)	Enzymatic activity (nmol/min)	Specific activity (nmol/min/mg protein)	Purification ratio
Soluble fraction	171.0	154	0.9	1.0
20-80% saturation ammonium sulfate fraction	127.0	150	1.2	1.3
Superdex 200	113.0	150	1.2	1.3
Ultrafiltration chromatogram	8.0	170	25.0	28.0
Hydrophobic chromatogram				
DEAE ion exchange chromatogram	2.8	180	64.0	71.0
Superdex 200 Ultrafiltration chromatogram 2nd time	0.3	210	700.0	778.0

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Fig. 5

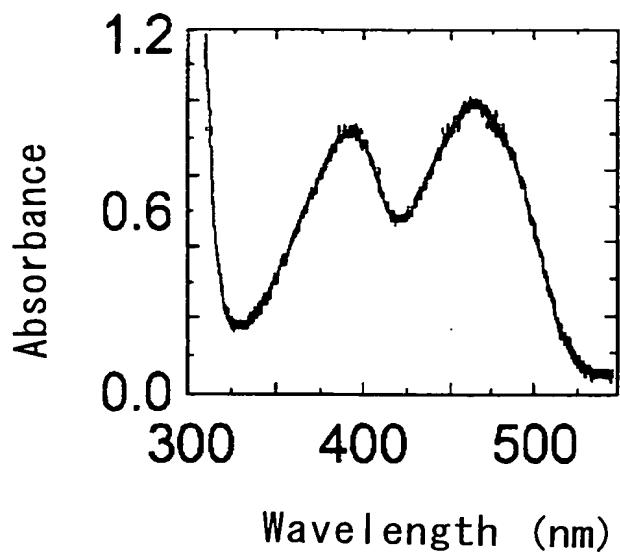
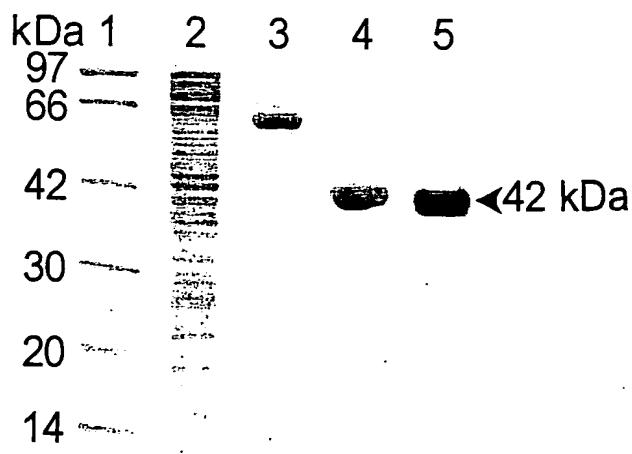


Fig. 6



1. Molecular weight marker protein
2. *E. coli* crude extract after transformation
3. Crude extract of *E. coli* expressing the recombinant TcOYE
4. Recombinant TcOYE collected by thrombin treatment
5. Purified standard of the recombinant TcOYE

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Fig. 7

Substrate specificity of reduction by the recombinant TcOYE

Substrate	Cofactor (10 μ M)	Km (μ M)	Vmax/specific activity (nmol/min/mg)
9, 11-endoperoxide PGH ₂	NADH	—	554
	NADPH	5.0	766
Hydrogen peroxide BHP ^a	NADPH	2.3	99
	NADPH	n. d.	282
Menadione	NADH	—	499
	NADPH	0.82	700
β -lapachone	NADH	0.17	650
	NADPH	—	433
4-nitroquinolin-N-oxide	NADH	—	759
	NADPH	9.5	1110
Nifurtimox	NADH	—	290
	NADPH	19.0	353
Phenazine methosulfate ^b	NADPH	10.4	235
Mevinolin ^c	NADH	n. d.	555
12-oxo-phytodienoic acid ^d	NADPH	n. d.	152
9-oxo ODE ^e	NADPH	n. d.	54
Econazole ^f	NADH	n. d.	43
Benznidazole	—	n. d.	N. D.
Miconazole ^g	—	n. d.	N. D.
Ketoconazole ^h	—	n. d.	N. D.
Crystal violet ⁱ	—	n. d.	N. D.
BHT ^j	—	n. d.	N. D.
BHA ^k	—	n. d.	N. D.

a: t-butyl hydroperoxide, b: 5-methyl-phenaziummethyl sulfate), c:
 2- β , 6 α -dimethyl-8 α -(2-methyl-1-oxo-butoxy)-mevinic acid lactone),
 d: 4-oxo-5 β -(2Z-pentenyl)-2-cyclopentene-1 β -octanoic acid),
 e: 9-oxo-10E, 12Z-octadecadienoic acid, f: 1-[2-([4-chlorophenyl]
 methoxy)-2-(2, 4-dichlorophenyl)ethyl-1H-imidazole], g: 1-[2, 4-
 dichloro β -([2, 4-dichlorobenzyl]-oxo)phenethyl] imidazole,
 h: cis-1-acetyl-4[4-[2-(2, 4-dichlorophenyl)-2-(1H-imidazol-1-
 yl-methyl)-1, 3-dioxolane-4-yl-methoxy]phenyl]piperazine],
 i: N-[4-[bis[4-(dimethylamino)-phenyl]methylene]-2, 5-
 cyclohexadiene-1-yl-iden-N-methyl-methane aluminum chloride,
 j: (2, 6di-tert-butyl-para-crezol), k: [2(3)-tert butyl-
 4-hydroxyanisole

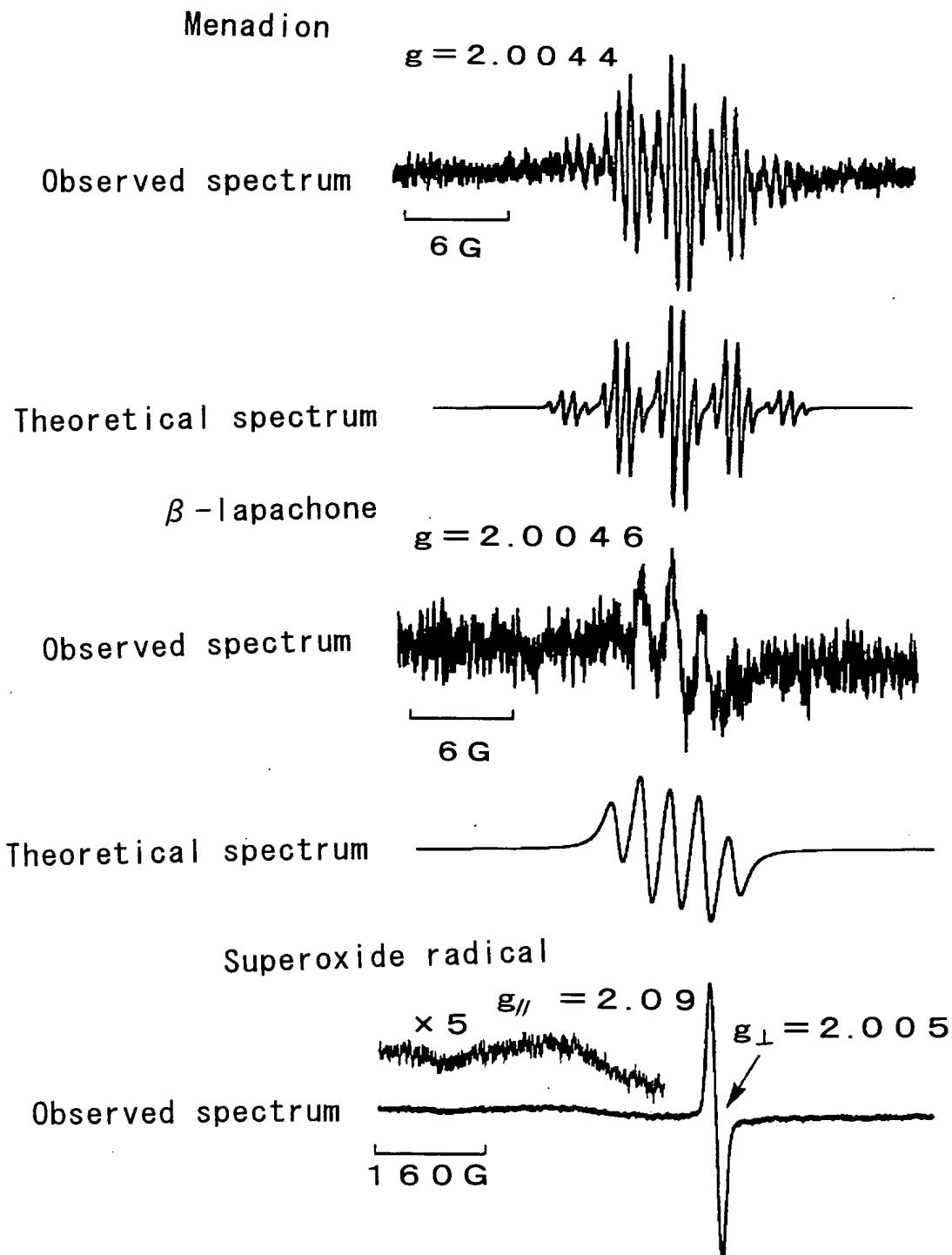
N. D. : not detected

n. d. : not measured

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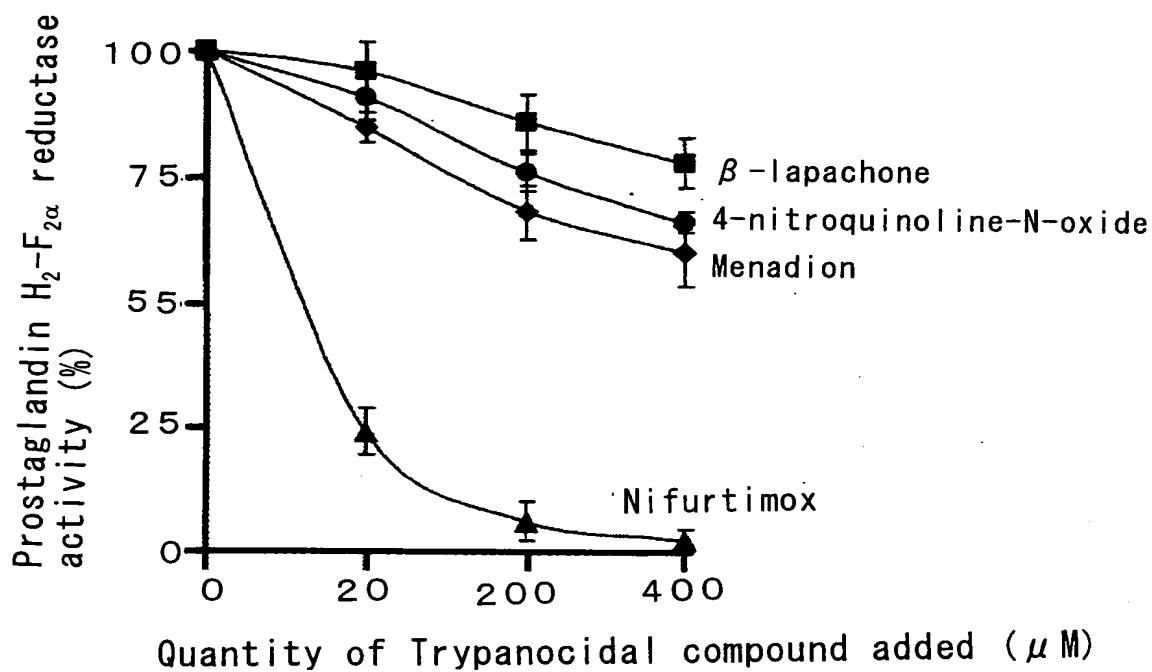
Fig. 8



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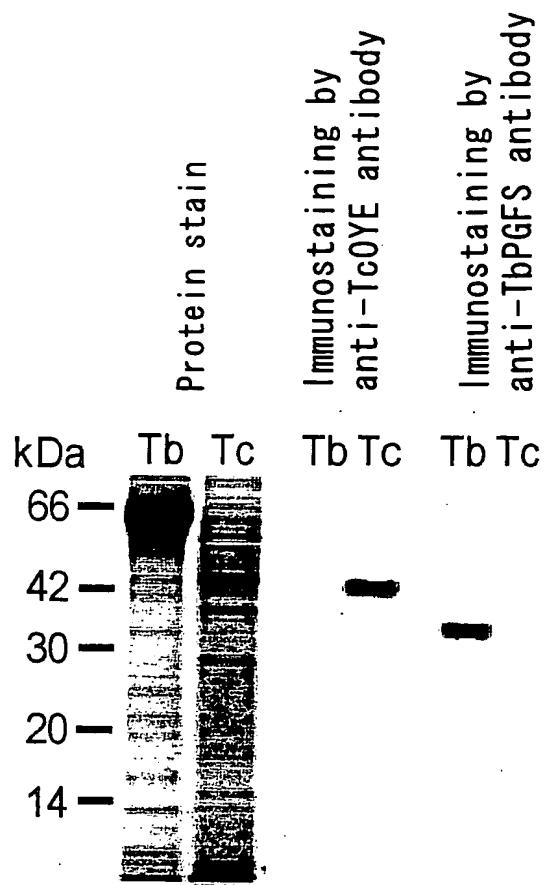
Fig. 9



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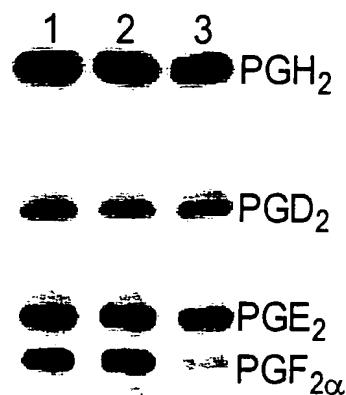
Fig. 10



Tb: Crude extract of *Trypanosoma brucei*

Tc: Crude extract of *Trypanosoma cruzi*

Fig. 11



1. Trypanosoma cruzi crude extract after reaction with control IgG
2. Trypanosoma cruzi crude extract after reaction with anti-TbPGFS antibody
3. Trypanosoma cruzi crude extract after reaction with anti-TcOYE antibody

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Fig. 12

Immunoprecipitation by the anti-Tc0YE antibody of the enzymatic activity to reduce menadion, β -lapachone, nifurtimox, 4-nitroquinoline-N-oxide in the crude extract of *Trypanosoma cruzi*

Sample	Persistent enzymatic activity (%)		
	Menadion	β -lapachone	Nifurtimox
4-nitroquinoline-N-oxide			
<i>Trypanosoma cruzi</i> extract after reaction with anti-Tc0YE antibody	100(± 2)	N. D.	N. D.
<i>Trypanosoma cruzi</i> extract after reaction with anti-TbPGFS antibody	98(± 8)	103(± 5)	100(± 5)
<i>Trypanosoma cruzi</i> extract after reaction with control bovine IgG	100(± 4)	100(± 10)	100(± 6)